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Ecological Morphology of *Abutilon theophrasti.*

BY LOUISE LUCKAN.

ABUTILON THEOPHRASTI, commonly known as velvet leaf, is an upright-growing mallow which may attain the height of six feet. It thrives at the edge of cultivated fields or in rich waste lands, such as lots, where it often occurs in dense patches.

During the summer of 1916 drought conditions prevailed for a month. The last rain of early summer fell about June 28 or 29, and the drought was not broken until the 26th of August. This meant that plant life would probably suffer for lack of water for at least a month. Many plants showed the effect of this lack of moisture very much, but *Abutilon theophrasti* remained fresh and green and attained its normal height. Here, then, an interesting problem offered itself as to what provisions the plant had made to withstand the drought.

The material for this study was collected on the south slope of the University campus. Both preserved and fresh material was studied. The standard histological methods were used and the drawings were made by the use of the camera lucida on the microscope and with the projectoscope.

ANATOMY.

The Leaf.

The leaves, borne on long, slender petioles, are roundish heart-shaped with an acuminate apex, and when full grown measure from four to five inches across. When the leaves are picked off they soon wither. During the hottest part of the day the edges of the leaf curl, but it requires only a short time for leaves to regain their turgor.

The shoots of young plants, each bearing from four to five leaves, were cut off, the cut surface sealed with wax and the shoots allowed to dry for an hour or more. These were then weighed separately and placed in a saturated atmosphere to see if they would regain their turgor or gain in weight. One set was left standing for forty hours, and then, after the leaf surfaces had been dried with filter paper, they were weighed. Those placed in the saturated atmosphere gained 15.11 per cent in weight, while those painted with distilled water and then placed in saturated atmosphere gained 18.95 per cent in weight. Another set was allowed to dry for three hours and was then brushed over with distilled water. The average of these tests showed a gain of 19.46 per cent in two hours. This shows that the plant has some power to absorb moisture from the atmosphere, especially if dew is present. Haberlandt ('14) gives two examples, *Convolvulus cneorum* and *Centaurea argentea*, where water is absorbed by hairs of the leaf. The wilted leaves gained 10 and 13 per cent, respectively, when immersed for twenty-four hours. In comparing the percentage gained and the conditions of the experiment, *Abutilon theophrasti* is much more efficient.

The leaf is dorsiventral, with the palisade tissue a single layer of cells on the upper side. Large thin-walled water-storage cells over the smaller veins interrupt this layer (fig. 1, *d*). Both surfaces of the leaf are thickly covered with four types of hairs, namely, two types of clothing hairs, one of which is stellate and the other an unbranched single-celled form, and two types of glandular hairs, one long and the other short. The stellate hairs, the most characteristic for the family, are the common clothing hairs, and these are found in great numbers and almost exclusively over the veins. The rays of the stellate hairs vary in number and length. They are united at their bases in the plane of the epidermis; and there being no stalk cells, these hairs do not rise far above the epidermal surface. The walls of the basal portion are relatively thick and contain pits (fig. 6). A ray cell (fig. 5) has the cavity diminishing toward the apex until in some cases it is entirely closed there. The walls of the rays show up as being composed of two layers—a thin outer cuticle imperfectly developed and a thick inner portion which the chloroiodide of zinc test gave evidence of being slightly infiltrated with cutin. The basal portion of these hairs stain a bright red with safranin, and yellow with chloroiodide of zinc, indicating cutinization, or possibly lignification; but give no color reaction with phloroglucin or ani-

line sulphate, and very little, if any, with Sudan III. In one test with the chloroiodide of zinc the inner portion of the cell wall of the basal region stained purple, indicating cellulose in the position shown in fig. 5. Some of these hairs showed nuclei situated in the basal part, but very little cytoplasm could be demonstrated.

Haberlandt, in his discussion of multicellular absorbing hairs, makes the statement: "If such a hairy covering is easily wetted and rapidly absorbs drops of water; if, further, the hairy leaf rapidly recovers its turgidity when immersed or besprinkled in the withered condition; if, finally, places for the entrance of water through the hairs are indicated by the presence of thin-walled basal cells with abundant protoplasmic contents, it may be safely assumed that the hairs in question serve to some extent for the absorption of water." The imperfect cutinization of the walls, above noted, would allow the water to pass to the inside of the hair. The structure of the stellate hairs, together with their occurrence only on the veins, and the amount of water absorbed by wilted leaves, as mentioned, lead me to believe that these hairs are water-absorbing hairs.

The other form of clothing hair is like a single ray of the stellate type; here the pits in the basal portion show up plainly (fig. 7).

The glandular hairs are of two kinds; one is long, being made up of from twelve to fifteen cells, while the other is only four to five cells in length. The basal cell of the long hair is enlarged (figs. 3 and 4). The cell just above it has its side walls cutinized throughout, so that it shows up as a bright-colored ring when stained with Sudan III or chloroiodide of zinc. The remaining cells of the hair are provided with a very thin cuticle. The apex is rounded out, and here drops of oil collect as it is secreted. The short glandular hair has its cells little differentiated, but the basal cell is slightly larger than the neighboring cells (fig. 8). These smaller hairs occur along the midrib and the larger veins, and Nestler ('99) says in some of the species of *Abutilon* they function as hydathodes, or hairs which excrete water. The hairs are advantageously situated for this, and as they contain living protoplasts they could possibly function in this way.

The stomata are not specialized, and are found in greater numbers on the lower epidermis, where they average 300 per square millimeter as compared with 250 on the upper surface.

The radial and outer walls of the epidermis are not especially thickened, but the inner wall of some of the epidermal cells becomes much thickened and mucilaginous and protrudes down into

the palisade tissue (fig. 2). The inner cell walls of the lower epidermis are not as thick as those of the upper, and only a few are mucilaginous. The mucilage may be so abundant, in varying degrees, that the inner wall at first glance might seem to be a second layer of epidermal cells, entirely filled with mucilage.

The mucilaginous deposits probably serve as a place of storage for water, absorbing a large amount when there is excess moisture in the leaf, and then gradually giving this up to the palisade tissue when the water supply runs low. I found no constant relation between these cells having the mucilaginous inner wall and the hairs, to indicate that water absorbed from the atmosphere by the hairs would be delivered to them for storage.

The midrib and the larger veins project prominently from the lower surface of the leaf, and the greater portion of this projection is composed of large thin-walled cells surrounding the vascular bundle (fig. 13). Kuntze ('91) describes mesophyll cells on the upper side of the vein, which break down to form large mucilage channels in many of the Malvaceæ. In the cross sections of the veins examined no channels were found, and only a single cell situated in this position showed mucilage (fig. 13, *m*). There are, however, on the upper surface a large number of epidermal cells, outlining the veins, which have a mucilaginous content (fig. 12). Therefore we may conclude that mesophyll cells of the midrib, as well as the mucilage cells, are good places for water storage, and are probably used as such.

The Stem.

The stem structure follows closely the type form of the dicotyledonous stem. The young stem is covered with both types of clothing and glandular hairs described, under the leaf. These hairs extend perpendicular to the surface, so that the young stem looks quite fuzzy. The epidermis of the stem has regular cells, the walls of which are not thickened to any great extent. The cuticle on the outer wall is very thin, and I found no cutinized layer which would aid in keeping in the water.

The cortex is made up of four regions—an outer thin-walled parenchyma, a collenchyma, a second thin-walled parenchyma, and the starch sheath (fig. 15). The outer thin-walled parenchyma is made up of three layers of radially enlongated cells. The collenchyma tissue is composed of three layers, forming a broken ring, with the gaps opposite the medullary rays. This arrangement allows the water to pass in and out to the epidermis more

readily than if it were to diffuse through the thickened walls of the collenchyma. The second layer of thin-walled parenchyma of the cortex is only one or two cells deep and is often crushed up against the collenchyma. The starch sheath is apparent, but the cells do not contain starch.

The bast strands of the pericycle are in groups, separated by thin-walled cells. The cell walls of the bast are lignified. Besides the bast of the pericycle there are several layers of secondary bast strands formed by the cambium and interspersed in alternate layers with the secondary leptome elements (figs. 14 and 15).

This growth develops a wedge tapering outward, so that in cross section the stem has wedges of bast and phloëm alternating with wedges of medullary ray, which taper in the opposite direction (fig. 14).

The medullary rays are only one to two cells wide in the xylem region of the stem, but where they extend out through the phloëm portion of the vascular bundle they broaden radially, so that here they are from four to six cells wide (fig. 15). The medullary rays of the stem do not contain large amounts of starch.

The phloëm is conspicuous, for there are between eight and ten layers of cells between the inner bast strands and the cambium.

Although this plant is an annual, the xylem of the stem seems to be made up of rings of growth due to alternate layers of tracheal tubes and xylem parenchyma (fig. 15).

The pith is permanent in the stem, increasing with the other tissues, so that in mature stems the pith has a diameter of one centimeter. In the younger pith the outer cells are filled with starch grains, and among these cells filled with starch are mucilage channels, or sometimes mucilage cells (fig. 14). These channels are formed by the cell walls becoming mucilaginous and some of them breaking down. They may be made of only one cell in the cross section, or several cells adjoining may break down so that the channels or pockets are of different sizes. The pith cells contain many calcium oxalate crystals, usually compound, which are very noticeable in the young cells, where they fill the cell cavity.

The Root.

The root is a long taproot with few secondary roots. The root resembles the stem in having large wedges of bast. The medullary rays, especially in the region of the xylem, are well filled with starch. Cork is present as an outer covering of the older portions of the root (fig. 16).

In the thin-walled parenchyma of the cortex, droplets of oil are of frequent occurrence, and these were discovered to be held in the stroma of spherical elaioplasts, which in unstained sections appear simply as highly refractive bodies. When treated with chloroform or ether for twenty-four hours and then tested with stains for oil, no oil was demonstrated, but irrigation on a slide with solvents of oil for ten to fifteen minutes did not cause all of the oil to disappear. Sections from celloidin material, which had been fixed with one per cent chromacetic acid, tested with Sudan III, alkanin and chloroiodide of zinc, showed these bodies as globular masses reacting as oil. Strasburger ('13) makes the statement that the chromacetic acid fixitive causes percipitation of the stroma of elaioplasts and renders the oil less soluble. This may explain oil remaining in the stroma of the elaioplasts through the celloidin process in which solvents of oil are employed. Safranin-hæmatoxylin stain and the three-color stain showed these bodies as purplish, spherical, reticulate structures. The elaioplasts in the cells of the root sometimes lie next to the nucleus, but a constant relation was not found. The cytoplasm in the older cells of the cortex is only a thin layer lining the wall, so that there the elaioplasts are against the wall; usually only one to a cell (figs. 17 and 18).

Beer ('09) in his work on *Gaillardia* sustains the theory of Wakker ('88) that the elaioplasts are formed by the aggregation and fusion of the leuocoplasts in the cell. His work seems to give conclusive evidence for this mode of formation in the plant studied. He found elaioplasts in different stages of development, and observed living cells in which the process was going on where cell divisions had long since ceased; but Politis ('11), in his work on the development of the elaioplasts in *Ornithogallum umbellatum* bulbs, maintains that they are formed by the cytoplasm, almost simultaneously with nuclear division. He states that the stroma of the elaioplasts stains with the nuclear stains, reacting like the nucleolus; and since they also are dissolved with nuclear solvents, he maintains the elaioplasts are of nuclear origin and gives the fact of the close association with the nucleus and the chemical reactions as basis for this conclusion.

In *Abutilon* I found elaioplasts were present in the actively growing parts, such as floral organs, growing apices of stem, young seedlings, barely sprouted seeds, and also in root sections in all stages of development. When the older parts of the stem were tested no elaioplasts could be demonstrated. The presence

of elaioplasts in the growing apex of the stem and the absence of them in the older stem seems to support Politis' theory that they are active only a short time and are then used up by the plant tissues, but the fact that the elaioplasts are present in the cells of the cortex of the old root is against this.

In regard to their origin, I am not able positively to confirm either theory. I find leucoplasts aggregated about the nucleus in the cells of the soaked seed, but I could not find intermediate steps, for it seems that in my material the elaioplasts do not form oil until they have attained a spherical form and ultimate size. Certainly the hairs of the *Gaillardia* studied by Beer are unusually favorable subjects for showing the course of elaioplast development.

Politis found the elaioplasts usually present in the epidermis of the floral organs, but their occurrence in other organs was inconstant. He says they are an active organ of the cell during growth. When active the elaioplasts have a rotating motion in the cell; when at rest the elaioplasts may assume various forms as a last phase of development. He found elaioplasts present in the following species of Malvacecæ: *Hibiscus syraicus, Althea rosea, Malva rotundifolia, M. sylvestris, Gossypium arboreum, Gothea cauliflora.*

MICROCHEMISTRY OF CELL CONTENTS.

The Leaf.

Leaves that had been in bright sunlight, brought in and tested for starch with iodine, showed the presence of starch. Similar leaves boiled in Fehling's solution showed crystals of cuprous oxide, indicating the presence of sugar in the form of monosaccharide.

Sections of leaves in Sudan III overnight showed palisade cells orange yellow, but not good demonstration of oil. The large glandular hairs showed droplets of bright orange red, indicating oil. Leaf sections soaked in chloroform and then tested with Sudan III did not give the red color reaction.

Volatile oil tests were made by collecting the distillate from leaf tissues on cover slips rinsed in chloroform, which were then treated with Sudan III, and iodine fumes. On cover slips without the distillate, but otherwise treated in the same way, no oil was demonstrated; on those with the distillate, however, a good positive test was shown, indicating the presence of volatile oil in the leaf.

Sections of leaves were treated with methylene blue as a mucilage test. The inner cell wall of some of the epidermal cells took the stain (figs. 1, 2*b*).

Sections of the leaf heated in Millon's reagent showed brick-red contents in the palisade and spongy parenchyma cells.

Sections treated with fresh ferric chloride solution gave no tannin reaction.

Compound crystals of calcium oxalate were demonstrated by the use of hydrochloric acid, in which the crystals disappeared without effervescence. These crystals were common in the border parenchyma of the large veins.

Stem.

Sections of older stems treated with iodine showed the presence of much starch in the outer cells of the pith and also in the medullary rays.

Fehling's solution used on sections of the stem showed cuprous oxide crystals, and these increased in number and size when sections were left in a incubator for several hours, thus indicating the possible presence of saccharose and glucosides in addition to glucose.

Young stems showed the presence of oil in elaioplasts, but in older portions of the stem the sections gave no oil reactions. The glandular hairs of the stem and the petiole have drops of oil collected at their apices. Sudan III stains these drops, collected on a slide, a bright red. When left on a slide on a 50° oven for twenty-four hours the oil did not disappear. It is thus probably fatty oil.

Protein tests were made with Millon's reagent, chloroiodide of zinc, and iodine, showing protein present only in the region of the phloëm and cambium. Methylene blue showed mucilage present in the pith and a very little in the cells of the cortex, and when fresh sections of the stem were placed on the slide and water applied to them, swelling and finally solution occurred in the secondary thickenings of the walls of some of the pith cells.

Calcium oxalate crystals are present in cells of the cortex and young pith.

Sections treated with ferric chloride gave no evidence of tannin.

Sections treated with saturated copper acetate solution for three weeks failed to show the presence of resin.

Alkaloid tests were made with potassium iodide and chloroiodide of zinc. Other sections soaked in alcoholic tartaric acid, a solvent of alkaloids, were compared with fresh sections. No brown precipitate was demonstrated, leading to the inference that no alkaloids were present.

Root.

Starch is present in the medullary rays. Methylene blue showed very little mucilage in the root. It was present only as a thin film in a few of the cell walls of the cortex. Fatty oil is present in the stroma of elaioplasts. Fresh sections soaked in solvents of oil, choloroform, ether and xylene, and then stained with Sudan III, showed the oil had been dissolved out and the stroma was partially collapsed. Tests for volatile oil were negative.

Protein tests are not decisive, because the cell walls of the lignified tissue became brick red with Millon's reagent. When these walls were tested with aniline sulphate and phloroglucin, they gave the lignin reaction.

Seed.

Sections of soaked seed treated with Sudan III and iodine showed a large amount of oil in the endosperm and also in the embryo.

Sections of seed treated with Millon's reagent turned red in the region of the endosperm and cotyledons, indicating the presence of protein as a reserve food. Other sections treated with iodine showed no starch was present.

Methylene blue showed no indications of mucilage.

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DESCRIPTION OF PLATES.

PLATE I.

FIG. 1. Section of leaf showing part of a large vein. × 300. *a*, upper epidermal cell; *b*, mucilaginous wall; *c*, palisade cell; *d*, water-storage cell; *e*, spongy parenchyma; *v*, vein; *f*, stellate hair; *g*, glandular hair.

FIG. 2. Cross section of leaf. × 285. *s*, stoma. The rest of the index is the same as in fig. 1.

FIG. 3. Long glandular hair. × 200. *a*, apex, *b*, basal cell; *c*, protoplast shrunken away from wall.

FIG. 4. Lower portion of long glandular hair from section of the petiole. × 325. *b*, basal cell; *d*, cutinized wall; *e*, cuticle.

FIG. 5. Longitudinal section through stellate hair, showing peculiar reaction in chloroidide of zinc; stipled, orange yellow; shaded, purple. × 325.

FIG. 6. Cross section of leaf showing stellate hair over vein; index same as fig. 1. × 325.

FIG. 7. Simple clothing hair from section of petiole. × 325.

FIG. 8. Short glandular hair. × 325. *a*, plasmolysed protoplast; *b*, basal cell.

PLATE II.

FIG. 9. Surface view of leaf, showing relation of hairs to the veins. × 200. *a*, long glandular hair; *b*, short glandular hair; *c*, stellate hair; *d*, stoma; *e*, vein.

FIG. 10. Tangential section of leaf. × 200. *a*, palisade cells; *b*, border parenchyma; *c*, tracheal tube.

FIG. 11. Tangential section of leaf, showing frequency of the mucilage cells. × 200. *r*, stoma; *h*, hair; *m*, mucilage.

FIG. 12. Tangential section of leaf, above a vein, showing frequency of mucilage cells there. × 200.

FIG. 13. Cross section of large vein. × 100. *a*, thin-walled cells; *p*, phloëm; *x*, xylem; *m*, mucilage.

PLATE III.

FIG. 14. Cross section of stem, showing the regions and the wedged effect of bast and medullary rays. × 15. *c*, cortex; *b*, bast; *p*, phloëm; *x*, xylem; *m*, mucilage; *me*, medullary ray; s, starch in pith.

FIG. 15. Section of stem, showing rings in xylem. × 150. *a*, epidermis; *b*, thin-walled parenchyma; *c*, collenchyma; *d*, second thin-walled collenchyma; *e*, starch sheath; *f*, bast fiber; *g*, leptome elements; *h*, phloëm; *i*, cambium; *j*, water tubes; *k*, wood parenchyma; *l*, pith; *m*, mucilage channels; *n*, medullary ray.

FIG. 16. Cross section of root. × 15. *c*, cork; *b*, bast; *p*, phloëm; *x*, xylem; *mr*, medullary ray; *r*, rootlets leaving the root.

FIG. 17. Cells of cortex of seedling root. × 325. *e*, elaioplasts.

FIG. 18. Cells of cortex of older root. × 325. Showing elaioplasts.

ABUTILON THEOPHRASTI.
Louise Luckan.

PLATE I.

ABUTILON THEOPHRASTI.
Louise Luckan.

PLATE II.

ABUTILON THEOPHRASTI.
Louise Luckan.
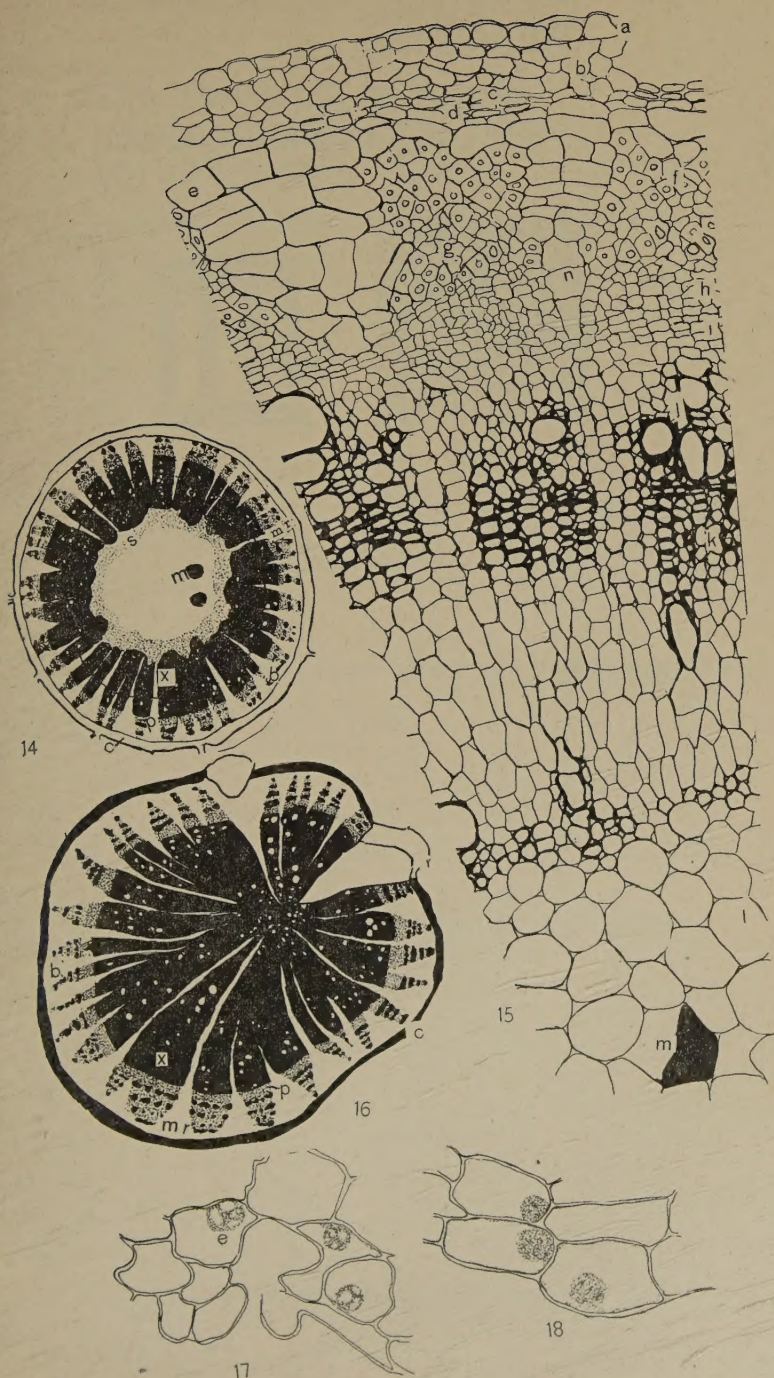
PLATE III.